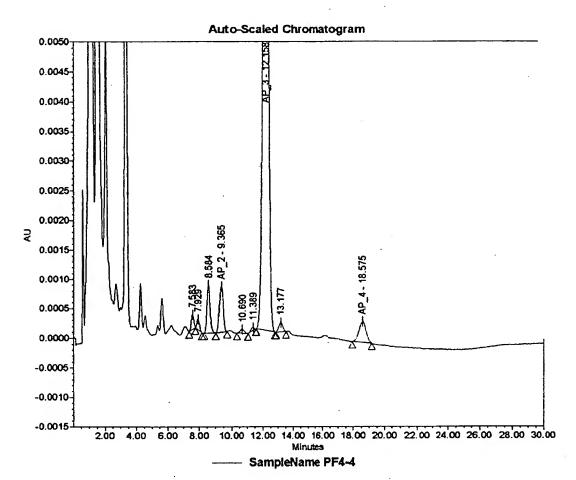


Ansamitocin P-3 (mg/L)

FIGURE 2

Lyophile (or frozen) culture Place on agar plate with medium CM4-1 28 °C, 5-7 days Pinpoint transfer to another CM4-1 agar plate 28 °C, 7 days Macerate each one colony in a screwed tube containing 10 glass bead (2mm dia.) and 2 mL H₂O Inoculate each 0.5 mL colony suspension to the seed (medium fill volume: 30 mL/250 mL flask) 28 °C, 48 hours shaker (throw: 70 mm; rotary speed: 220 rpm) Transfer 1.0 mL grown seed to fermentation medium (fill volume: 20 mL/250 mL flask) 28 °C, 8 days shaker (throw: 70 mm; rotary speed: 220 rpm) Harvest Use 4.5 mL ethanol (95%) to extract each 0.5 mL whole broth Mix (extract for 10 min on a vortex mixer (SMI model 2600) Centrifuge Supernatant for hplc assay

FIGURE 3



G4 AP-3

Sample PF4-4 PF4-4	Component	7.583 7.929	Area 2809	Height 251	Amount
PF4-4				251	
		7,929			
PF4-4	 		2168	178	
		8.584	11415	818	
PF4-4	AP_2	9.365	12486	785	14.062
PF4-4		10.690	1354	71	
PF4-4		11.389	821	56	
PF4-4	AP_3	12.158	345473	17000	389.092
PF4-4		13.177	3001	160	
PF4-4	AP_4	18.575	10938	357	12.319
1	PF4-4 PF4-4 PF4-4	PF4-4 PF4-4 AP_3 PF4-4	PF4-4 10.690 PF4-4 11.389 PF4-4 AP_3 12.158 PF4-4 13.177	PF4-4 10.690 1354 PF4-4 11.389 821 PF4-4 AP_3 12.158 345473 PF4-4 13.177 3001	PF4-4 10.690 1354 71 PF4-4 11.389 821 56 PF4-4 AP_3 12.158 345473 17000 PF4-4 13.177 3001 160